# ESTIMATING LEAF WATER STATUS FROM VIS-NIR REFLECTANCE AND TRANSMITTANCE

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## **ABSTRACT**

Remotely sensing the water status of plant canopies remains a long term goal of remote sensing research. Established approaches involve measurements in the thermal infrared and the 900-2000nm reflective infrared. Less popular UV-visible-NIR techniques presumably deserve research attention, because photochemical changes linked to plant water status manifest spectral light scattering and absorption changes. Here we monitored the visible and NIR light reflected from the leaf interior as well as the leaf transmittance as the relative water content of corn (Zea mays L.) leaves decreased. Our results highlight the importance of both scattering effects and effects due to absorption by leaf pigments.

*Index Terms*— leaf relative water content, RWC, leaf reflectance, leaf transmittance

# 1. INTRODUCTION

Remotely sensing the water status of plants and the water content of canopies remain long term goals of remote sensing research [1]. Estimates of canopy water status commonly involve measurements in the 900nm – 2000nm or thermal infrared portions of the optical spectrum and the Crop Water Stress Index (CWSI) [2] [3] - or its improvement, the Water Deficit Index (WDI) [4] - or the Equivalent Water Thickness (EWT) [1][5].

CWSI, the first widely adopted remote sensing plant water stress index, is tied to plant physiology, its principle advantage. It provides indication whether plant stomata are open or closed based upon the principles of evaporative cooling and the foliage radiant temperature relative to the surrounding air temperature. CWSI theory assumes a closed canopy having wall-to-wall vegetation and no soil visible from above the canopy. Moran et al. [4] modified and extended the CWSI theory, proposing the Water Deficit Index (WDI) in order to account for effects due to the bare soil often visible between rows of plants in agricultural crop

canopies and between the vegetation patches common to dry environments.

Limitations affect CWSI/WDI popularity and applicability. First, it does not work well in regions with high humidity where evaporative cooling effects may be limited. Second, its use requires estimates of the water vapor pressure deficit during the remote sensing over-flight; weather stations that provide such estimates are not always helpfully located next to the canopy. Third, once water stressed plant canopy stomata close, CWSI/WDI will indicate that the canopy is indeed water stressed but not how close that canopy is to its permanent wilting point (when plants die). Finally, its use requires analysis of calibrated, atmospherically corrected, thermal infrared data. Such data are not always readily available.

Estimates of EWT equate the water in a canopy to a hypothetical horizontal layer of water [5]. EWT estimates depend upon remotely sensed measurements in the reflective infrared, 900nm-2000nm where light absorption by water varies with wavelength. Such estimates are easily made, a key EWT advantage compared to CWSI/DWI, but depend upon the physics of water-light interaction, not upon plant physiology - which is the key limitation with EWT. Without *a priori* information, it is not possible to determine from estimates of canopy EWT if plants displaying a specific EWT are, for example, satiated or at their permanent wilting point or somewhere in between. However, it should be noted that canopy water status often can be inferred when EWT estimates are interpreted based upon other remote sensing results.

In addition to CWSI/WDI and EWT estimators, plant canopy water status may be estimated using the Photochemical Reflectance Index (PRI) and fluorescence [6] [7], although neither enjoys the popularity nor the robustness of the CWSI/WDI and EWT approaches. A key issue with both is that any observed change in PRI or fluorescence has not one but many potential causes [7]. That is, if the canopy PRI value or the amount of fluorescent light emitted by a canopy changes, the potential cause of

that change could be a change in plant canopy water status — or incident light level, nutrient availability, ambient temperature, wind speed and direction or other factor that affects the plant photochemistry. However, the additional information needed to attribute one cause to an observed change is sometimes available; the case of carefully managed, irrigated, agricultural plant canopies provides one example.

Visual indicators closely linked to plant water status have been reported. For example, leaves, when water stressed, may droop or curl into a tube (canopy architectural changes) and appear gray (a spectral light scattering change) compared to fully hydrated leaves that typically appear turgid and vivid green. However, robust, widely accepted remotely sensible metrics that exploit such visual indicators for estimating canopy water status are not available.

In prior research we reported a linear relationship between the light reflected by the interiors of individual corn leaves measured *in vivo* and the leaf relative water content (RWC),  $R^2 = 0.77$  [8]. In our recent research [9], we reported a linear relationship between the ratio

and the leaf relative water content, RWC.

Here we report results of our continuing search for robust estimators of plant canopy water status based upon remotely sensible measurements of the uv, visible and near infrared spectral regions. Such estimators potentially would provide access to the plant hydrological photochemistry that occurs in the 300-800 nm spectrum.

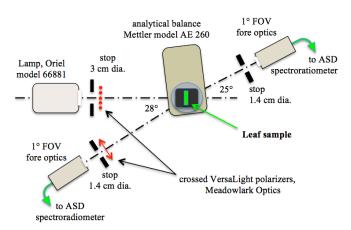


Fig. 1. Plan view of the experimental setup. The reflectance, transmittance and weight of the leaf sample were monitored as the leaf lost water. During dry down, the leaf's water status changed from fully hydrated to near the permanent wilting point.

#### 2. METHODS

Within 2 hr of sunset, we selected 5 sweet corn (*Zea mays* L.) plants in the 7-8 leaf stage from a large field. We lifted each plant with its root ball largely intact and placed each one in a large clear plastic bag with 500 ml of water. The bags were sealed and kept overnight at room temperature to rehydrate the leaves.

The next morning in the lab, when a leaf sample was needed for purposes of measuring the leaf bidirectional reflectance and bidirectional transmittance, we first completed as quickly as possible the following sequence: open the bag, cut an upper fully expanded leaf from the plant, blot the now fully hydrated leaf dry, trim the leaf in order to select a 4.5 cm x 4.5 cm leaf sample, mount the leaf segment in the sample holder and, place it on the pan of the analytical balance, Mettler model AE 260 (Fig. 1).

We illuminated the leaf, Fig. 1, with a collimated beam of white light provided by a current controlled lamp, Oriel model 6681, and immediately began collecting spectral data and sample weights with the aid of the analytical balance and two Analytical Spectral Devices spectroradiometers. The leaf sample, initially fully hydrated at 100% RWC, rapidly began losing water when exposed to the light beam.

Data collection continued for typically 0.75-1.5 hours until we estimated the leaf sample RWC was less than 65% (approximately the permanent wilting point for corn).

We later dried the leaf samples in a 65°C oven for 2 days, cooled the leaf samples and estimated RWC for a specific leaf weight as

$$RWC = \frac{[leaf\ wt.] - [leaf\ dry\ wt.]}{[leaf\ fully\ hydrated\ wt.] - [leaf\ dry\ wt.]}$$

We calibrated the spectra using a multi-step procedure involving observation of Spectralon<sup>TM</sup> by both reflectance and transmittance spectrometers and observation of opal glass by the transmittance spectrometer. The crossed polarizers eliminated the light reflected by the leaf surface, allowing the reflectance spectrometer to observe only the light reflected by the leaf interior. We collected spectral data as the leaf dried and later calculated the leaf interior reflectance, R, and the leaf transmittance, T.

The leaf interior reflectance  $R(RWC,\lambda)$  and the leaf transmittance  $T(RWC,\lambda)$  were normalized at each wavelength  $\lambda$  by  $R(RWC\sim0.97,\lambda)$  and  $T(RWC\sim0.97,\lambda)$  when RWC was approximately 0.97.

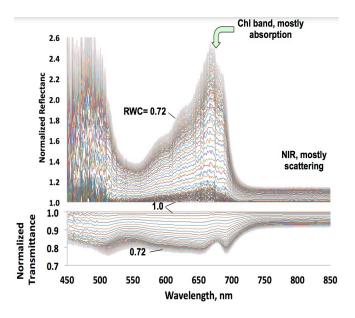


Fig. 2. Results for one sweet corn leaf. The leaf interior reflectance R and leaf transmittance T, normalized by R(RWC~0.97, $\lambda$ ) and T(RWC~0.97, $\lambda$ ), display variation with wavelength related to leaf pigments. As the leaf dries and RWC decreases from 1.0 to 0.72, nearly the permanent wilting point of the leaf, normalized R increases and normalized T decreases.

# 3. RESULTS AND DISCUSSION

Our results, Fig. 2, show that both the leaf normalized interior reflectance R and leaf normalized transmittance T display no variation with wavelength in the 800-850nm spectral region (the NIR) where leaf pigments little interact with incident light. Conversely, between 500-700nm (the visible) both R and T show pronounced wavelength variation that is typically associated with leaf pigments. Note also that normalized R and normalized T values for

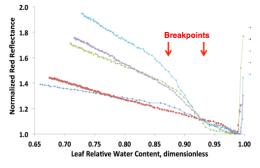


Fig. 3. Normalized R in the chlorophyll absorption band around 680nm increased monotonically for RWC<0.97 for five sweet corn leaf samples. The transient effects for 0.97<RWC<1.0 are attributable to chlorophyll fluorescence effects in these dark adapted leaves.

0.97<RWC<1.0 are not displayed in Fig. 2 but are displayed in Figs. 3 and 4. This choice means that transient effects in the 450-550nm and 670-780nm spectral regions do not appear in Fig. 2. Such transient effects in R and T have been linked, for example, to fluorescence effects and other pigment effects in dark adapted leaves.

Our results, Fig. 3, suggest there are 'breakpoints' in the normalized R of four of the five sweet corn leaves measured in the red chlorophyll absorption region as RWC decreased. The appearance of the two breakpoints suggests sequential domination of chlorophyll degradation by three processes as water in the leaf decreases.

Fig. 4 shows that the R<sup>2</sup> for three line segments regressed to the topmost curve in Fig. 3 is high, 0.94 or greater. No similar breakpoints were evident in the normalized R v. RWC response curves (not shown) for the green and NIR spectral regions. All of this further supports the possibility that the apparent decrease in chlorophyll pigment related absorption is sequentially dominated by three processes related to the decrease in water in the leaf.

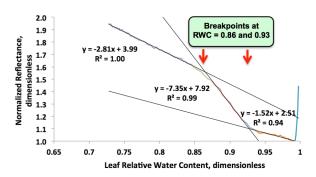


Fig. 4. Line segments were fit to the topmost curve in Fig. 3.

In [9] we proposed that the optical light scattering changes occurring as a leaf dries could be understood with reference to a simple model based upon the leaf cellular structure – upon the changes to the cell wall and cell membrane of a cell as it loses water. When fully hydrated, cells are turgid, pressurized like plump grapes. As they lose water, the internal pressure in the cells goes to zero - 'zero turgor' and cells become flaccid. Beyond zero turgor as the amount of water in each cell continues to decrease, changes to the cell wall and cell membrane become evident as the cell membrane pulls away from the cell wall. Eventually, if dehydration continues, cells collapse and appear like wrinkled raisins. From optics the cell surface area per unit volume increases as the cell's water status changes from zero turgor to its permanent wilting point (when cells do not recover).

All this means that as the leaf water status changes from fully hydrated to zero turgor little change in cellular light scattering should occur because little cellular structural changes occur. But from zero turgor to the permanent wilting point, the amount of light scattered by the cell and thus by the leaf should increase, leaf transmittances should decrease and the amount of light reflected by the leaf interior should increase. We reported results, Fig. 5, [9] that support this simple model. This model only considers light scattering; it ignores cellular light absorption changes due, for example, to changes, as cells lose water, in the molecular configuration of pigments.

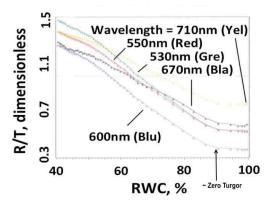


Fig. 5. The ratio leaf interior reflectance/leaf transmittance increases linearly as relative water content (RWC) decreases below zero turgor, approximately 90%. (Reproduced from [9]

The pronounced pigment absorption effects evident in our results, Fig. 2, show that the simple model [9] can not adequately describe the changes to R and T that we report here. Another anomaly involves two breakpoints, Figs. 3 and 4, compare to the one breakpoint evident in Fig. 5.

## 4. CONCLUSIONS

Our previously proposed conceptual model [9] includes no absorption, only scattering and can not adequately represent our results reported here, which display pronounced absorption effects related to chlorophyll pigment.

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